The Report Committee for Marcus Andrew Ebels
Certifies that this is the approved version of the following report:

The Use of Plant Growth Promoting Bacteria as 'Bio-Fertilizers': Crop Inoculation to Reduce Agrochemical Devastation

APPROVED BY
SUPERVISING COMMITTEE:

Supervisor: _____
R. Malcolm Brown, Jr.

_____
Mona Cynthia Mehdy

The Use of Plant Growth Promoting Bacteria as 'Bio-Fertilizers': Crop Inoculation to Reduce Agrochemical Devastation

by

Marcus Andrew Ebels, B.A.

Report

Presented to the Faculty of the Graduate School of

The University of Texas at Austin

in Partial Fulfillment

of the Requirements

for the Degree of

Master of Arts

The University of Texas at Austin

August 2015

Dedication

This report is dedicated to Dr. Dennis Jackson of Eastern Michigan University, without whom I would not have taken the chance on grad school and would still be burdened with a great deal of student debt. You were right about deferment. Also your enthusiasm for teaching, which motivated me as an undergrad, now inspires my future career aspirations. Parenchyma!

Acknowledgements

I would like to thank all the professors at UT who have taught me so much, both in lecture and on how to lecture. Dr. Brown, for giving me the opportunity to move to Austin and try out the wild world of academia. It has helped me grow in ways I never would have guessed and I feel all the more prepared for whatever lay ahead. Dr. Mehdy, for being willing to lend an ear and showing much needed kindness when I was having a hard time. Dr. Hawkes, for your lectures which had a considerable impact on my interests and by going out of your way to help me pursue them. Gustavo Ochoa, for listening to me gripe and giving sound advice. Dr. Latham, for helping me to lead discussions and get invaluable experience, your way with the students is definitely a goal I will keep in mind. And Dr. Omelon, for giving me the chance to showcase my talents. It has been an honor to take part in this program.

Abstract

The Use of Plant Growth Promoting Bacteria as 'Bio-Fertilizers': Crop Inoculation to Reduce Agrochemical Devastation

Marcus Andrew Ebels, M.A.

The University of Texas at Austin, 2015

Supervisor: R. Malcolm Brown, Jr.

Plants harbor a diverse community of microbes which may be beneficial or detrimental to their health. Among these microbes, plant growth promoting bacteria (PGPB) perform functions which may provide the host plant with nutrients and phytohormones or help manage environmental stress and antagonize pathogens. The use of PGPB as inoculants in agriculture is often referred to as bio-fertilization and can reduce the need for chemical fertilizers, pesticides and herbicides through the various activities provided by the bacteria. In application of PGPB, inoculation may be through the coating of seeds and treatment of furrows with a free liquid culture. It may alternatively involve the encapsulation of PGPB in biodegradable beads which reduce the loss of cells to environmental constraints and competition/predation by other soil microbes while prolonging PGPB activity and benefit to the host crop. Bio-fertilizers are currently in use but further development is needed in testing and production to make a variety of products more available for use with a wider range of crops.

Table of Contents

Introduction

The relationship between microbes and the organisms they inhabit involves a wide array of interactions which may be harmful, helpful or of little to no consequence to the host. The human digestive tract hosts a diverse community of microbes, known as the gut microbiome, which carries out functions such as the synthesis of vitamins, degradation of complex polysaccharides and regulation of the host immune system (van Olden et al., 2015). In return, microbes are provided with a nutrient rich environment in which to thrive. This familiar example is used to illustrate mutualism, a relationship between different organisms in which both benefit. Advances in identification of microbes and the processes they carry out in mutualistic interactions have shed light on their necessity and the true cost of their loss. For instance, more fully understanding the role of the gut microbiome exposes damaging effects of antibiotics not previously realized. As all multicellular organisms host microbes, greater knowledge is needed in the cultivation of functionally diverse microbial communities.

Similar to animal systems, the microbiome of plants gains a nutrient rich environment for growth. External microbes are also considered a part of the microbiome of an organism (Such as bacteria on the skin) and may be mutualists as they can still supply benefits to their host. For plants, much exchange with external microbiota takes place below ground through the activity of the roots. The rhizosphere is the area of soil in close enough contact with roots to be exposed to root exudates, and is delineated for its

rich microbial life. The exudates released into the rhizosphere affect microbial activity and the compounds released can alter the community, partially through attracting motile microbes, resulting in beneficial or detrimental effects for the plant (Berg and Smalla, 2009; Drake et al., 2013; Liu et al., 2015b). This zone is heavy with inter-organismal crosstalk, which can result in alterations in the physiological activities of both the plant and microbes. For instance, salicylic acid, a phytohormone which plays a role in pathogen resistance through coordinated plant cell death, has been found to affect the virulence of plant pathogens, reducing their ability to successfully invade tissues (Xu et al., 2015). Conversely, pathogenic microbes are capable of targeting salicylic acid biosynthesis through the release of virulence factors, reducing its concentration in tissues and exudates (Tanaka, Han and Kahmann, 2015). In this way, the rhizosphere serves as an interface between the plant and its associated microbiome which can determine not only the health of the plant but its growth and survival. Understanding the underlying activity of these interactions holds the potential for applications in agriculture and environmental restoration.

Within the host plant, microbes referred to as endophytes may be found colonizing tissue. These internal symbionts may be parasites or mutualists as endophyte only applies to physical location and not activity. In the case of endophytic bacteria, colonization of tissues may result in the formation of specialized structures, such as root nodules (Del Papa et al., 1999) or coralloid roots (Costa, Paulsrud and Lindblad, 1999), or endophytes may have no effect on morphology, still living between or within cells

(Dong et al., 1994). Mutualistic plant-bacterial relationships such as these are thought to have most often arisen from parasitic interactions (Sachs et al., 2014). Distinction being made between colonization by mutualists and pathogen invasion is vague as they essentially describe the same process. Pathogens' prolonged association with plants resulting in a loss of virulence is also evidenced in the close evolutionary relatedness of some endophytic mutualists to parasites (Blakney and Patten, 2011). An evolutionary simulation using a parasitic bacterium carrying a symbiotic plasmid serves as another example of endophyte evolutionary history. A symbiont plasmid common to rhizobia was transferred to the pathogen and it was subsequently found that conversion to a nonpathogenic, nodule-forming endophyte was able to be accomplished with a loss-of-function mutation to a single virulence regulatory gene (Marchetti, 2010). As plasmids can be transferred between bacteria through conjugation with the same efficiency as the methods used here, this study demonstrates the relative ease with which this conversion could take place in nature. Despite these contradictory origins, the transformation to mutualism is thought to be evolutionarily stable (Sachs et al. 2014). Pathogens and mutualists evolve to have some level of dependence on their host. However these relationships also affect the fitness of the host and since these interactions have undoubtedly been affecting plant survival and fecundity, it seems understandable that the microbiome of plants has become an integral part of growth and productivity.

Endophytic bacteria and those present in the rhizosphere which perform functions that stimulate production by the associated host plant are considered Plant Growth

Promoting Bacteria (PGPB). As these bacteria are mutualists, these benefits are provided in exchange for metabolites, whether in the apoplast or in root exudates. PGPB represent a subset of the plant microbiome which can directly affect plants' chances of survival given environmental constraints and infectious disease.

The ability of plants to cultivate microbial communities, and the microbes' effect on growth, can be used in aiding difficulties faced in agriculture (Bakker et al., 2012). The practice of crop rotation uses legumes' microbiota to increase the soil nitrogen content for the following year's crop (Bahl and Pasricha, 2000). Intercropping fields has been found to improve growth and yield, wherein the microbial community cultivated by one plant enhances the growth of a different, concurrently planted crop (Debenport et al., 2015; Hernandez et al., 2015). These are two examples of sustainable agriculture which can reduce the need for agrochemical practices, referring to the use of chemical fertilizers and pesticides (Bakker et al., 2012). The use of fertilizers on a large scale has broad environmental effects such as eutrophication in bodies of water and soil acidification (Menesguen and Dion, 2009; Vassilev et al., 2015). Chemical fertilizer has also been shown to reduce the abundance, diversity and functionality of bacterial soil communities as compared with organic fertilizer (Chauhdry et al., 2009). The use of chemical pesticides and herbicides reduce the plant growth promoting activity of soil bacteria (Ahemad and Khan, 2011; Ahemad and Khan 2012; Abbas et al., 2015) along with the diversity of bacterial communities (Tan et al. 2012; Wang et al. 2013). The effects of toxins in chemical pesticides also have ecological impacts on mutualistic animal

relationships with plants, such as the destruction of primary pollinators (Tavares et al., 2015)

Amending farming practices which are detrimental to the microbiome of plants and focusing more on enhancing PGPB growth and activity for the benefit of crops has the potential to reduce costs and adverse environmental side effects. Though the name is somewhat ambiguous, bio-fertilizer is often used to describe an inoculum for agricultural uses which can improve plant productivity and growth by application of microbes such as PGPB. This review focuses on the various activities of PGPB and how different activities carried out by bacteria can be applied to increase the growth and productivity of crops including advancements in methods of inoculation.

PGPB: Host Benefits and Conditional Uses

PGPB are a diverse group of bacteria able to provide a variety of advantages to their host. This allows for application of particular strains based on crop species, environmental conditions or pathogen type. Overlap exists in these traits wherein one activity by PGPB may benefit in multiple ways or a single strain may exhibit multiple beneficial traits. Despite this difficulty, this information is intended to describe the individual advantages of some of the most prominent PGPB traits and showcase examples of their use in inoculations.

PHOSPHATE SOLUBILIZATION

Phosphorous is a major limiting nutrient for plant growth as much of the content of soil is in the form of insoluble phosphates. Both the organic and inorganic forms of insoluble phosphate (P) can be converted by PGPB into soluble forms which are able to be absorbed by plants. Organic immobilized P is solubilized by the secretion of degradative enzymes (Rodriguez et al., 2006) while inorganic sources of P, such as calcium phosphate, is solubilized by the secretion of organic acids (Xiao, Fang and Chi, 2015).

Inoculation with known P-solubilizing bacteria has been found to increase plant growth both in lab studies (Bautista-Cruz et al., 2015) and in field studies (Saleem, Arshad and Yaseen, 2013; Viruel et al., 2015). The addition of rock phosphate or phosphorite, along with P-solubilizing bacteria yields increased growth and production in maize and wheat as compared with inoculation alone or the addition of chemical fertilizer

(diammonium phosphate) (Kaur and Reddy, 2015). In this study the addition of a P rich mineral substrate for P-solubilizing PGPB increased the uptake by plants to a greater extent than fertilizer. In a similar study, the use of rock phosphate alone increased populations of P-solubilizing bacteria in soil and produced growth and yield results similar to chemical fertilizer (triple super phosphate) in field trials (Ndungu-Magiroi et al., 2015). While bacterial populations were increased by rock phosphate, the addition of chemical fertilizer reduced the occurrence of the same bacteria in the soil.

In an attempt to increase the activity of one P-solubilizing isolate, Liu et al. (2015a) used genetic recombination of another solubilization gene. The strain was previously shown to solubilize inorganic phosphate using organic acid but the introduced gene encoded for a phytase from a *Bacillus* strain. Phytase is able to cleave an organic source of P (phytic acid), making a soluble compound. Thus the transformed strain was found to utilize both inorganic and organic phosphorus sources. This was confirmed by conversion of phytic acid by the transformant and increased levels of soluble phosphorous in the medium of the transformed strain as compared with its parental strain. Lab trials using maize confirmed that growth rate was significantly greater in corn seedlings inoculated with the transformed strain as compared to the parental strain resulting in increased dry shoot weight. This strain could potentially be used as an effective inoculant in bio-fertilizer treatments.

NITROGEN FIXATION

Nitrogen (N) is another major limiting nutrient for plant growth as it is mainly in its inert gaseous form, dinitrogen. The ability to reduce dinitrogen to organic N in the form of ammonia is limited to bacteria and archaea and catalyzed by the enzyme complex nitrogenase (Pedraza, 2008). Rhizobia, a family of PGPB, colonize the roots of legumes providing organic N and receiving metabolites from the host. This may be one of the most studied PGPB relationships and may also be responsible for the first forms of bio-fertilizers, with commercial inoculants reaching as far back as the late 19th century (Bashan, 1998). Despite this history of use, methods of inoculation are still actively investigated (Hynes et al., 1995; Bogino et al., 2011). Increasing root nodule function has been accomplished by co-inoculation of rhizobia with other endophytic root colonizing bacteria (Yadegari et al., 2010; Subramanian et al., 2015a) resulting in higher N accumulation by the host.

Another N-fixing endophytic PGPB, *Gluconacetobacter diazotrophicus*, was originally isolated from sugarcane, but has been found to colonize other crop species including rice, sweet potato, pineapple and finger millet (Saravanan et al., 2008). *G. diazotrophicus* is an example of an endophyte which is able to live throughout the tissues of the host plant, in the apoplast. Growth and survival of sugarcane seedlings is increased with inoculation of *G. diazotrophicus* (de la Cruz, Bird and Isulat, 2012) and plants treated with the PGPB show greater mass accumulation associated with higher N content compared with un-inoculated plants (Singh et al., 2012). In field trials comparing inoculation combined with fertilizer use, inoculation with *G. diazotrophicus* along with

50% fertilizer addition resulted in a 4.53% in yield over the full application of fertilizer alone (Anitha and Thangaraju, 2011). Not only does *G. diazotrophicus* reduce the need for fertilizer but the addition of chemical fertilizers (ammonium sulfate and calcium nitrate) in high doses reduces both the population and N-fixing activity of *G.* diazotrophicus *in vivo* (Medeiros, Polidoro and Reis, 2006) This is thought to be the result of reduced nitrogenase expression by *G diazotrophicus* resulting in an inability to compete with other soil microbes and/or inhibition of colonization by the host.

While N-fixing endophytes can serve as bio-fertilizer inoculants, they are confined in use by host specificity and micro-aerobic environments as oxygen can interfere with nitrogenase function. An approach to the issue of host specificity undertaken by Barney et al. (2015) used the free living rhizospheric bacterium *Azotobacter vinelandii*. Not only free living but able to fix nitrogen in aerobic environments, *A. vinelandii* does not provide this organic N to potential hosts as excretion from the cell is minimized by enzymatic and transport activity. In this study, random transposon mutagenesis was used to increase N excretion from *A. vinelandii*. The insertion of transposons in the gene *amtB*, encoding an ammonium membrane transporter, resulted in significantly higher levels of ammonia in the medium as compared with the wild type strain. The loss-of-function mutation disabled the transporters ability to retrieve lost ammonia from the surroundings. This increase in excreted organic N was capable of supporting the growth of co-cultured algal species allowing the *amtB* mutants to be identified. Though levels of excreted ammonia were still

relatively low, this work suggests that the free living *A. vinelandii* could potentially be used in bio-fertilization with soil applications to any given crop species.

PHYTOHORMONE REGULATION

Endophytes may produce or alter the host's biosynthesis of phytohormones, compounds which regulate plant growth and development. It is important to mention that phytohormone production is a common characteristic of both parasitic and mutualistic plant endophytes and can result in the formation of pathogenic (Aragón et al., 2014; Gohlke and Deeken, 2014) and mutualistic associated structures (Fedorova et al., 1992). However not all endophytes which produce phytohormones form structures and may instead have broader effects on overall organ morphology (e.g. increased shoot length) or may increase the biosynthesis of specific metabolites (Wang et al., 2015b). It should be noted that these effects can improve plant growth but their individual effects in the use of bio-fertilizers can be difficult to parse out as their production is often carried out with other PGP activities such as nutrient acquisition and pathogen antagonism.

Indole-3-acetic acid (IAA) is major compound in the auxin family of phytohormones. Auxins are important in the regulation and coordination of plant growth and IAA production has been identified in many PGPB (Ona et al., 2005; Rajkumar et al., 2005; Sethia et al., 2015). Research on tumor formation such as galls and nodules highlights the importance of IAA in tumorogenesis in plant organs. The IAA synthetic pathway involved often reflects the bacterial relationship to its host. There are five separate biosynthetic pathways of IAA synthesis from tryptophan and certain pathways

are associated with different endophytic activities. Phytopathogens mainly synthesize IAA through the indole-3-acetamide pathway, which is associated with the formation of tumor like structures, while PGPB mainly utilize the indole-3-pyruvate pathway, enhancing plant growth (Patten, Blakney and Coulson, 2013; Lin, Chang and Lin, 2015).

In an example which focuses on the individual effect of bacterial phytohormone related activity, a *Bacillus* strain isolated from the roots of Arabidopsis was discovered to enhance plant growth by altering the host's expression of IAA related genes rather than producing IAA of bacterial origin (Wang et al. 2015a). Plants inoculated with the *Bacillus* isolate exhibited increased growth of lateral roots and root hairs while the extension of the primary root was limited. These alterations of root morphology resulted in a significant increase in shoot fresh weight and leaf area. This change was partially the product of induced transcription of IAA biosynthesis genes but also involved changes in the expression of auxin transporter genes. This illustrates a more indirect mode of phytohormone associated activity which still has a substantial effect on plant morphology and physiology.

Cytokinins are another family of growth inducing phytohormones, and their ratio to auxin has been found to determine lateral versus apical shoot growth and the development of organs and tissues in growing plant embryos (Schmulling, 2002; Muller et al., 2015). The production of cytokinins has been identified in PGPB with very direct benefit for the microbes producing it. A rhizospheric isolated PGPB strain of *Bacillus subtilis* which produces zeatin type cytokinins was used to investigate the role of this

phytohormone in mutualistic plant-microbe interactions (Arkhipova et al., 2005). When inoculated in the rhizosphere of wheat, this strain was found to increase exudation of amino acids from roots by 30%, the same effect as treatment of roots with direct application of zeatin. This may seem less than advantageous for the host plant, but the PGPB strain provided other growth promoting effects besides phytohormone production, exhibited by higher nitrogen and chlorophyll content of inoculated plants (Kudoyarova et al. 2015).

Nitric oxide (NO) is another compound involved in plant/microbe interactions. Though the designation of NO as a phytohormone is not universal, it is a gaseous signaling molecule with a wide range of developmental and physiological effects, on par with ethylene (Shapiro, 2005; Jin et al. 2011). It is included with phytohormones and will be referred to as such as it acts in concert with other phytohormones as a part of processes such as senescence, root development, seed germination (Sanz et al., 2015; Kopyra and Gwóźdź, 2004) and response to abiotic and biotic stresses (Shi et al., 2012). Plants are able to synthesize their own NO, but PGPB also contribute NO production to these responses (Yamasaki, 2000).

In plants, levels of NO and IAA are correlated and therefore both are involved in similar developmental processes (e.g. root hair formation) (Pagnussat et al., 2002). In bacteria, NO regulates traits such as virulence, mobility and biofilm formation. When bacterial IAA is produced concurrently with NO, the same correlation between the phytohormones is exhibited as in plants (Koul, Adholeya and Kochar, 2014). In a study

using a strain of *Azospirillum brasilense* which produces both NO and IAA, compounds which affected the production or accumulation of either NO or IAA were found to simultaneously affect levels of the other phytohormone (Koul et al., 2015). Similar effects on root morphology were seen in inoculated plants as with PGPB-IAA production alone (increased lateral root hair formation/growth) but the additional production of NO had a synergistic effect with IAA, further improving root hair development.

ABIOTIC STRESS TOLERANCE

Under condition of low water availability or high salinity, there is an increase in photorespiration resulting in the buildup of reactive oxygen species (ROS) which damage proteins and DNA. Phytohormones regulate responses to abiotic stress through stomatal closure, senescence and abscission (Miller et al., 2010). Auxins, ACC deaminase and cytokinins have been identified as bacterially produced compounds responsible for enhancing drought tolerance of the plant host (Liu et al., 2013; Belimov et al., 2015). The alleviation of environmental stress highlights the potential use of bio-fertilizers in enhancing growth in adverse conditions.

Further investigation into the mechanisms of conferring drought tolerance by endophytic PGPB uncovered changes in genetic expression of the host. The *B. subtilis* strain used in the study was confirmed to perform PGP activities such as P-solubilization and IAA production (Gagne-Bourgue et al., 2013). Using previously identified sequences in the plant species *Brachypodium distachyon* for genes involved in water limited plant response, Gagne-Bourgue et al. (2015) found that expression was significantly higher

when inoculated plants were grown in drought stressed conditions as compared with un-inoculated plants. Sequences included two dehydrins, enzymes responsible for mitigating osmotic stress, and a transcription factor acting upstream of their expression. It was also found that the genome of inoculated plants showed increased methylation and that these epigenetic changes may prime the host for drought tolerance. The aforementioned genetic changes are not directly tied to PGPB phytohormones (i.e. IAA) but this concurrent activity may prove to be involved in this process as plant growth was increased and time to maturation was also accelerated.

NO is similar to other phytohormones in regulating drought responses such as stomatal closure, but it's also an antioxidant, capable of reacting with ROS forming H_2O_2 and thereby reducing damage cell components. NO enhances the activity of H_2O_2-scavenging enzymes as well, furthering the full conversion of NO to O_2 (Siddiqui, Al-Whaibi and Basalah, 2010). By inoculating wheat with a strain of *Lactobacillus plantarum*, capable of producing NO when provided the precursor L-arginine, drought tolerance was discovered to be the product of increased antioxidant capacity in tissue extracts of inoculated plants. The catalytic activity of tissue extracts when adding H_2O_2 was also higher than un-inoculated control plants (Yarullina et al., 2014). This highlights the underlying mechanisms of drought tolerance and the ability to protect plants through inoculation.

Working downstream of ROS buildup and NO signaling pathways, similar effects on plant stress responses are seen by acting on ethylene. ROS act as signaling molecules

themselves, and their accumulation mediates ethylene responses to stress in plants (Xia et al., 2015). The enzyme 1-aminocyclopropane-1-carboxylate (ACC) deaminase reduces levels of an ethylene precursor, thereby reducing ethylene buildup which if prolonged can inhibit growth. This growth inhibition through ethylene buildup can result in greater detriment to plant productivity than the initial salt stress (Madhaiyan, Poonguzhali and Sa, 2007). ACC deaminase production by endophytic PGPB is able to mitigate stress related inhibition to a variety of environmental conditions (Grichko and Glick, 2001; de Souza et al., 2015). For instance, high salinity limits growth in seedlings and is linked to ROS and ethylene buildup (Glick, Penrose and Li, 1998; Hussain et al., 2015). Inoculated plants with ACC deaminase producing PGPB show increased growth of roots and shoots when grown in saline soils owing to a reduction in the ethylene precursor ACC (Ali, Charles and Glick, 2014; Zafar-ul-Hye, Farooq and Hussain, 2015). In another example, the use of transformed strains of cold tolerant PGPB were used to assess the role of ACC deaminase in chilling stress of tomato (Subramanian et al., 2015b). Cold tolerant strains were transformed with *acdS*, the gene encoding ACC deaminase, and seeds of tomato were inoculated before germination. The effect of ACC deaminase expression by PGPB was apparent as inoculated plants with prolonged exposure to 10˚C showed increased dry weight and reduced levels of ethylene emission compared to un-inoculated plants.

Heavy metal contaminated soils cause phytotoxicity and limit the growth and survival of plants (Goncharova et al., 2010; Matraszek and Hawrylak-Nowak, 2010). Metal resistant endophytic and rhizospheric bacteria are able to be isolated from polluted

areas and inoculation of plants with metal resistant PGPB has been shown to reduce the phytotoxic effects of contaminated soils (Tank and Saraf, 2009; Yang et al., 2013; Pereira, Barbosa and Castro, 2015). Phytoremediation, the removal of contaminants from soil using absorption by plants, can be enhanced by activity of metal resistant PGPB (Aung et al., 2015; Ma et al., 2015). A major factor in tolerance to heavy metals contributed by PGPB is the production of siderophores. As a family of chelators, siderophores have high affinity for metal ions and increase their bio-availability through solubilization of compounds such as iron hydroxides (Boukhalfa and Crumbliss, 2002). In the context of PGP, this solubilization alone has the capacity to increase plant growth in limited conditions (Radziki et al., 2013). But as a part of abiotic stress tolerance, bacteria isolated from heavy metal contaminated areas have been found to have very active siderophore production (Kumar et al., 2015) and the metal chelating ability allows for greater plant tolerance and up take of heavy metals (Jeong, Moon and Nam, 2014; Nagata et al., 2015).

PHYTOPATHOGEN ANTAGONISM

Although more related to pesticide than fertilizer, PGPB inoculation is also able to enhance growth through reduced damage of organs and tissues by pathogens. Antagonism is therefore an indirect PGPB activity which is especially useful for crops which are devastated by infectious disease (Nakaew, Rangjaroen and Sungthong; 2015). Cavendish banana trees, being genetically identical clones, are struck very hard by fungal infection, leaving entire crops destroyed. PGPB isolated from the rhizosphere of banana

has shown strong antifungal capacity and broth preparations from cultures rivaled copper based pesticides in antifungal activity while having no negative effects on other rhizospheric PGPB (Shaikh et al., 2014).

Antagonistic activity of PGPB takes several forms, and because more than one may be carried out by the same bacterium, the effect of individual compounds can be unclear. These include the production of antibiotics, phytohormones, siderophores, volatiles and lytic enzymes (Zhang et al., 2014; Khabbaz et al., 2015). These compounds can act in unison carrying out different functions in order to suppress pathogenic invasion.

Chitinase is a lytic enzyme specific to the suppression of fungi, degrading the main cell wall component of the pathogen. As these enzymes need to be excreted into the extracellular environment, they are very stable in a wide range of temperature and pH (Karthik, Binod and Pandey, 2015). Interestingly, lytic enzyme production and excretion has been found to vary based on the exposure to different fungal species, exhibiting the bacterium's ability to maximize degradative capacity (Zhao et al., 2013). Inoculation of crops with chitinase producing PGPB has been shown to suppress fungal pathogens (Han et al., 2015). In one study, the activity of a chitinase producing strain of PGPB was found to be increased by providing chitin in the inoculation medium (Yandigeri et al., 2015). The increased production of chitinase through chitin supplementation resulted in enhanced fungal suppression.

Alongside providing stress tolerance, siderophores can act as agents of pathogen suppression. Though less direct than lytic enzyme secretion, the production of siderophores limits the availability of iron for fungal pathogens as the bacterial chelators have higher affinity than those of fungal origin. This sequestration by PGPB results in suppression of disease through nutrient deficiency (Maindad et al., 2014; Sulochana et al., 2014). Siderophores have also been found to reduce the toxicity of the pathogenic fungal metabolite fusaric acid. Members of the fungal genus *Fusarium* produce this compound and it is toxic to both bacteria and plants, causing wilt and rot diseases in the latter. The sequestration of iron by siderophores reduces its toxicity, as fusaric acid requires the binding of iron as a part of its mode of action. In a study by Ruiz, Bernar and Jung (2015), bacterial siderophore production increased with the addition of fusaric acid and PGPB mutants unable to produce the ligands showed a loss of resistance to the toxin.

Inoculations with PGPB can be used to suppress bacterial infections of plants. This may involve the use of antibiotic compounds and volatiles (Chung et al., 2015). Other means of suppression can be in the form of pathogen signal disruption. Quorum-sensing, the ability of a bacterial population to sense its density through the concentration of an extracellular signal, is a main factor leading to increased virulence and pathogenesis. The act of reducing virulence through interfering with this signaling pathway, known as quorum-quenching, is a different method of ceasing pathogenesis which does not involve attacking or limiting the growth of a potential pathogen. Endophytic PGPB of cannabis employ quorum-quenching, reducing the production of

quorum-sensing signal molecules and thereby protecting the host from infection (Kusari et al., 2014). As resistance to antibiotics becomes more prevalent, this method of disease suppression holds potential in both plants and animals.

As related to the topic of supplementation of pesticide use with PGPB inoculation, herbivory can also be potentially targeted with the use of bio-fertilizers. As inoculation with endophytes can protect all or particular tissues of the host, these treatments can be very effective and make reapplication unnecessary. Production of endophytic PGPB toxins can increase the mortality of insect herbivores. Cry1Ac toxin, produced by *Bacillus thuringiensis* delays larval growth and reduces pupal weight and adult emergence of corn borer moths (Ma et al., 2008). For herbivores which target root tissues, rhizospheric inoculations can also be effective. In a study using rhizospheric PGPB, the damage to roots of tomato by nematodes was reduced (Mahgoob and El-Tayeb, 2010). This was again accomplished through toxin production resulting in reduced nematode population density. Treatments were found to be effective up to a period of 30 days as the bacterial population of the rhizosphere declined after this period.

VOLATILE ORGANIC COMPOUNDS

Plants mediate many microbial interactions through the production of volatile organic compounds (VOCs). Along with metabolites such as carbohydrates and amino acids, VOCs are thought to play a role in the cultivation of the rhizosphere microbial community. Along with metabolizing VOCs of plant origin, bacteria take part in this inter-organismal mediation by producing VOCs of their own (Junker and Tholl, 2013).

VOCs are signaling molecules but are also capable of enhancing plant growth. VOCs gaseous nature means they can take affect even in tissues of the host which are not in close proximity to the bacterial source (Park et al., 2015). Some of these compounds are able to both enhance growth and antagonize pathogens, working both directly and indirectly to promote the growth of the host plant (Hernández-León et al., 2015).

Inocula: Methods and Applications

In lab trials, the use of sterilized soil allows for inoculation and the colonization of test plants with little to no interference. Though some previously discussed cases involved field testing, most utilized sterility in determining the individual contribution of PGPB inoculants. In open, unsterilized soil, inoculated bacteria have to compete with already established bacterial communities (which are often more adapted to soil conditions) and are under the additional complication of environmental variables such as soil type and plant species constituting the rhizosphere. Exposure to predation by protozoans may also account for a significant loss of inoculated microbes. These difficulties often result in the decline of bacterial populations following inoculation (Bashan, 1998). Given the potential for failure of the PGPB to provide sustained benefits, the tailoring of inocula to both the strain of bacterium and the environmental conditions is a critical part of bio-fertilizer application. This section will attempt to cover advances in types of inoculation and the advantages of each.

FREE PGPB INOCULATION

In many lab trials of inoculants, seed coat sterilization and inoculation is the method imposed for PGPB treatment. This method primes the seed allowing the germinating seedling to become colonized. Studies using this method in field trials have also seen significant increases in growth of the host and increased yield (Díaz-Zorita and Fernández-Canigia, 2009). Carriers used for inocula in seed coat treatment may be liquid or solid. Comparisons of different bacterial carrier materials including sand, sawdust,

vermiculite and liquid waste from the cosmetic industry were used to test the ability of these materials to successfully transfer PGPB after prolonged periods of inactivity (Maheshwari et al., 2015). These carriers were treated with strains of PGPB and mixed in a solution containing the thickening agent carboxymethyl cellulose in order to be used as seed coating inoculants. Seeds were treated with the different carriers for a period of up to a year after the initial inoculation of the carrier materials. Vermiculite was found to contain the highest amount of viable cells after a year and also had the greatest increase in growth and yield. This capacity to harbor viable endospores was attributed to the high water holding capacity of the mineral. These carriers were chosen for their cheap cost and potential use in broad field treatments. More testing would be required to verify field use but the results were promising as even cosmetic waste proved to be an efficient carrier of PGPB after a year of storage.

In-furrow inoculation refers to treating the soil with liquid inoculations before planting. This method used in legumes has been shown to allow inoculated endophytic bacteria a greater advantage in colonizing the host, out competing wild nodule-forming competitors (Bogino et al., 2011). These treatments were compared with seed coating treatments, in which the native endophytes composed the bacteria found in nodules in all trials. This suggests that in order to establish a desired endophyte, in-furrow inoculation is advantageous. In another study comparing in-furrow and seed inoculation of legumes, no significant increase in host growth was exhibited between the two applications. However when seeds were treated with fungicide, in-furrow treatment resulted in

significantly increased plant growth and yield as compared with seed coating inoculation (Zilli et al., 2010).

In-furrow, liquid inoculations have the advantage of being uniform, easily handled and bringing about a quick response in the inoculated plant. However, because inoculated PGPB are exposed to the environment and microorganisms of the soil, these effects can be limited. Even when using sterilized soil, as in the previously mentioned study of nematode suppression by PGPB (Mahgoob and El-Tayeb, 2010), effects were seen to decrease after the soil was treated, as inoculated bacteria could not be established with the crop.

ENCAPSULATED PGPB INOCULATION

When inoculating with endophytic PGPB, open treatments which consist of spraying soil or seed coats with culture media are effective as the environment sought out by the bacterium is ultimately within the plant. However these same treatments used with PGPB which inhabit the rhizosphere become limited as, depending on the soil and level of competition therein, the inoculated microbes may not survive long and offer little help to the plant. In order to protect PGPB from the challenges faced in fields and furrows, encapsulation of inocula in synthetic polymers serves as a barrier, giving the PGPB with nutrients to sustain them and separate them from the external soil environment. The polymers used are organic in nature, and as they are degraded, the inoculant PGPB are released into the rhizosphere. This release is therefore gradual and can extend the inoculation process over a greater span of time, allowing more benefit to be provided to

the host crop (Bashan, 1998). The shelf life of encapsulated PGPB is much longer than free inoculants, with viable cells being able to be produced after 14 years of storage (Bashan and Gonzalez, 1999). The use of encapsulation is effective for use in general PGP such as nutrient provision or more specific applications such as pathogen antagonism and abiotic stress tolerance (Wu et al., 2011).

Alginate is a material well studied in the use of encapsulation of PGPB. It is a polysaccharide formed by β-1,4-linkages of L-glucuronic and D-mannuronic acid and is produced by macroalgae and some bacteria. It has a high water absorbing capacity making it ideal for harboring microorganisms. Beads of alginate can be freeze-dried after bacterial encapsulation and PGPB are able to be reactivated when the beads absorb water. In lab trials using sterile soil, alginate encapsulated bacteria used in inoculation were found to perform as well in enhancing growth as in-furrow style treatments using direct liquid inoculant on the soil (Rekha et al., 2007). Encapsulation using alginate can be enriched with the addition of nutrient sources, which may increase activity or longevity of the internal PGPB. The addition of humic acid to the alginate bead composite showed significantly enhanced growth of the host as compared with PGPB inoculation alone (Young et al., 2006). This was attributed to the PGPB being stimulated with extra nutrients as soon as they were reactivated by the absorption of water by the alginate beads.

The release of PGPB from capsules is initiated by degradation of the bead material by both the encapsulated bacteria and external bacteria in the environment.

However, in the initial absorption of water by freeze-dried capsule, swelling of the beads accounts for a considerable loss of PGPB cells into the soil. Soil factors that affect absorption include pH, temperature and salt content. Blending alginate with starch and clay (bentonite) affects the release of cells through altering the ability of the beads to swell on contact with moisture. Starch has many hydroxide groups which interact with water molecules increasing the swelling of beads, corresponding with the percentage of starch added to the composite. This results in more cells released. The addition of clay reduces swelling and cell release, likely because of mechanical strengthening of the beads (Wu et al., 2012). The increased release of cells exhausts the beads internal PGPB population more quickly, therefore depending on the application this may be undesirable. Clay is capable of prolonging the release of cells, with a lessened initial release. The addition of other materials to the beads also reduces cost, as less alginate is needed in a composite (He et al., 2015).

Polycaprolactone (PCL) is another polymer of interest in PGPB encapsulation. PCL is a biodegradable plastic, more difficult to degrade for microbes than alginate but not to the point of causing environmental build up over time. Similar to alginate, the addition of starch and clay (montorillonite) can be used to amend the release of cells from the bead, with starch and clay increasing and decreasing cell release, respectively. However, PCL is hydrophobic and the release is accounted for by microbial degradation, especially by the internal PGPB. The addition of starch reduces the strength of PCL and increases permeability while clay increases mechanical strength (Wu, Wu and Chang,

2007). PCL can be used in a multiphase encapsulation method to offer internal PGPB extra protection against external competitors and predation. Enclosing cells suspended in alginate with PCL has no negative impacts on cell viability and can increase the encapsulated PGPB biosynthesis of desired compounds. By encapsulating lactic acid bacteria in multiphase beads with PCL, production of lactic acid production was nine times higher than in free inoculants. Lactic acid is antagonistic to nematodes and the capsules were found to be effective in suppression of nematode parasitization (Takei et al., 2008).

The hydrophobic nature of biodegradable plastics creates difficulty when making a composite bead, hence the multiphase encapsulation using alginate. The first phase, alginate encapsulation, can be carried out with an aqueous solution and the second, PCL encasement, with an oil based solution. In another example of a hydrophobic polymer, polybutylene succinate (PBSU), the addition of starch to the material composite was accomplished with the grafting of an acrylic acid to the polymer, increasing hydrophilicity while retaining the degradative qualities of the polymer (Wu, 2008). As before, the addition of starch could subsequently be used to decrease the strength of the beads and increase biodegradability/cell release.

CONSORTIA

Multiple bacterial species may be used in the same inoculant to increase PGP. These are referred to as consortia and their combined activity can have greater effects on plant growth through additional PGPB activities or through synergistic effects on a

particular activity (Yu et al., 2012). Inoculation with two strains of endophytic bacteria can improve colonization of both strains of PGPB (Maheshwari et al., 2010). However this is not always the case as competition and antagonism can potentially exist among consortia. When inoculating sugarcane with five species of endophytic PGPB, the population of individual species within the host varied with which bacteria were present among the five species. Though most of the species colonization improved with the addition of others, *Gluconacetobacter diazotrophicus* populations were highest when inoculated alone (Oliveira, 2009).

Encapsulation may also be used with this method to effectively combine PGPB activities. This includes examples of indirect methods of enhancing plant growth such as abiotic stress and pathogen antagonism (Guo et al., 2012) and direct enhancement through increasing availability of limiting nutrients (Mishra et al., 2011) with evident synergistic effects given the environmental conditions. This suggests the potential of broader uses of inoculants when incorporating multiple strains. Certain considerations must be taken into account when encapsulating more than one strain as the capsule environment may need to be adjusted to accommodate both. If strains are of the same species this consideration is often unnecessary (Bashan, 1998).

Other biosynthetic activities could be used in consortia to the advantage of PGPB survival. *Azotobacter* is a genus which includes species of PGPB able to fix nitrogen and produce phytohormones. These PGPB are also capable of producing an alginate exopolysaccharide (EPS). EPSs form biofilms which encase bacteria in the same method

as encapsulating materials, protecting from predation and environmental conditions. They also serve as adhesive material for bacteria to the surface of the host plant. This protective barrier could offer the same shelter to other inoculated PGPB, prolonging their survival in the soil and beneficial activity (Gauri, Mandel and Pati, 2012).

Though this review has focused solely on growth promotion by bacteria, it is important for this topic to discuss the advantages of inoculation of PGPB with mutualistic fungi of plants. Fungi offer the decomposition of organic compounds in the soil, providing nutrients to the host (Vassilev et al, 2015). This degradation can also provide energy for high cost of biosynthetic activities of PGPB, such as nitrogen fixation (El-Katatny, 2010). Fungi may also carry out other PGP activities such as phytohormone production. A strain of endo*hyphal* bacteria, living within an endophytic fungal host, was found to increase IAA production by the fungus, enhancing growth of the all-encompassing plant host. Interestingly, the bacteria do not produce IAA on their own, but only enhance fungal synthesis. This interaction is somewhat complex, i.e. a bacterium within a fungus enhancing growth within a plant, but an excellent illustration of the complexity of inter-microbial relationships and the potential use of consortia of both fungi and bacteria (Hoffman et al., 2013). Of course, antagonism is also a possibility when forming consortia between different microbial organisms. In fact environment variables such as soil pH can determine whether the relationship between fungus and PGPB is cooperative or antagonistic (Sousa et al., 2015).

Conclusions

The practical use of PGPB as bio-fertilizers offers many advantages to crops but many of the applications discussed previously require more testing and the ability to be produced on a large scale in order for the average farmer to consider their use. Seed treatments and liquid inoculations offer quick results but lack longevity. Encapsulation can offer the prolonged benefit of PGPB inoculation but costs of producing encapsulated bacteria remain high. And all potential inocula, whether single strain or consortium, require background testing and tailoring to the strains and crops intended for cultivation (Tomic, Milivojevic and Pesakovic, 2015). However most of this information is not difficult to collect, especially if the product of interest is in trial use. BioGro is one example of bio-fertilizers which are currently available. Testing on inoculant products is still being carried out to increase efficiency with different crop varietals and reduce cost through timing of applications (Hien et al., 2014).

Inoculation of soy bean with rhizobia and wheat/maize inoculation with *Azospirillum* have been reported to take place in Central and South America on an estimated 25 million and 500,000 hectares, respectively. Though vast areas are affected, the introduction of PGPB has been found to have much less of an effect on soil community composition than environmental filtering (e.g. moisture and temperature) and host filtering (e.g. root exudates) (Castro-Sowinski et al., 2007). This evidence fits with the Baas-Becking hypothesis, that microbes are universal, everything is everywhere but the environment selects. Some previously discussed studies included the use of transgenic

microorganisms the field use of which is more complicated. Although the same rules of filtering apply to transgenic bacteria, their effect on community structure in the soil is of greater concern, especially considering conjugation and the ability of bacteria to horizontally transfer genetic material. This entails tracking diversity and community structure along with the prevalence of the transgene following the introduction of the PGPB strain (Lynch et al., 2004). Gathering information about this would be needed before any field trials of potential transgenic inoculant strains.

Until more products are available for a wider range of crops, reduction in the use of chemical fertilizers through the use of organic manure and crop-waste fertilizers can help increase microbial diversity. Diverse microbial communities are also more functionally diverse and therefore offer more benefits to crops without inoculation. Soil microbial communities play an integral role in nutrient cycling and the transfer of energy through ecosystems. As chemical fertilizers reduce diversity and inhibit activity of soil microbes along with causing environmental damage, any reduction in their use builds towards a more functional soil community and healthier crops.

Glossary

Chelation – The binding of an organic compound with a metal ion.

Endophyte – Organism which lives within plant tissues.

Mutualism – Relationship between two organisms in which both are benefitted.

Parasitism – Relationship between two organisms in which one benefits and the other is negatively affected.

Phytopathogen – Disease causing microbe, specifically of plants.

PGPB – Plant growth promoting bacteria, a mutualistic part of the plant microbiome which may be endophytic or external and stimulates growth in the host. PGP is also used herein as shorthand for plant growth promotion.

Quorum sensing – The excretion of a signaling molecule by all individuals of a bacterial population which allows each individual to sense the density of the population. Higher concentrations of the signal therefore bring about responses by the entire population unanimously.

Siderophore – Low molecular weight organic chelators which have a very high binding specificity for Fe(III) ions, however the binding of other metal ions has also been identified.

Symbiotic – Relationship in which the organisms involved are intimately associated, e.g. one living within the other.

Rhizosphere – Soil which surrounds the surface of a root and is exposed to the compounds which it extrudes.

Vermiculite – A naturally occurring mineral with insulating properties.

Virulence – The relative capacity of a pathogen to cause disease.

References

Abbas Z, Akmal M, Khan KS, Fayyaz ul H. 2015. Impacts of long-term application of buctril super (bromoxynil) herbicide on microbial population, enzymes activity, nitrate nitrogen, Olsen-P and total organic carbon in soil. Archives of Agronomy and Soil Science. May;61:627-644.

Ahemad M, Khan MS. 2011. Toxicological assessment of selective pesticides towards plant growth promoting activities of phosphate solubilizing pseudomonas aeruginosa. Acta Microbiologica Et Immunologica Hungarica. Sep;58:169-187.

Ahemad M, Khan MS. 2012. Evaluation of plant-growth-promoting activities of rhizobacterium Pseudomonas putida under herbicide stress. Annals of Microbiology. Dec;62:1531-1540.

Ali S, Charles TC, Glick BR. 2014. Amelioration of high salinity stress damage by plant growth-promoting bacterial endophytes that contain ACC deaminase. Plant Physiology and Biochemistry. Jul;80:160-167.

Anitha KG, Thangaraju M. 2011. Gluconacetobacter diazotrophicus to influence leaf N and enhance cane yield by increased colonization under reduced N fertilization in sugarcane. Research on Crops. Aug;12:453-461.

Aragon IM, Perez-Martinez I, Moreno-Perez A, Cerezo M, Ramos C. 2014. New insights into the role of indole-3-acetic acid in the virulence of Pseudomonas savastanoi pv. savastanoi. Fems Microbiology Letters. Jul;356:184-192.

Arkhipova TN, Veselov SU, Melentiev AI, Martynenko EV, Kudoyarova GR. 2005. Ability of bacterium Bacillus subtilis to produce cytokinins and to influence the growth and endogenous hormone content of lettuce plants. Plant and Soil. May;272:201-209.

Aung HP, Djedidi S, Oo AZ, Aye YS, Yokoyama T, Suzuki S, Sekimoto H, Bellingrath-Kimura SD. 2015. Growth and Cs-137 uptake of four Brassica species influenced by inoculation with a plant growth-promoting rhizobacterium Bacillus pumilus in three contaminated farmlands in Fukushima prefecture, Japan. Science of the Total Environment. Jul;521:261-269.

Bahl GS, Pasricha NS. 2000. N-utilization by maize (Zea mays L.) as influenced by crop rotation and field pea (Pisum sativum L.) residue management. Soil Use and Management. Sep;16:230-231.

Bakker MG, Manter DK, Sheflin AM, Weir TL, Vivanco JM. 2012. Harnessing the rhizosphere microbiome through plant breeding and agricultural management. Plant and Soil. Nov;360:1-13.

Barney BM, Eberhart LJ, Ohlert JM, Knutson CM, Plunkett MH. 2015. Gene Deletions Resulting in Increased Nitrogen Release by Azotobacter vinelandii: Application of a Novel Nitrogen Biosensor. Applied and Environmental Microbiology. Jul;81:4316-4328.

Bashan Y. 1998. Inoculants of plant growth-promoting bacteria for use in agriculture. Biotechnology Advances. Jul;16:729-770.

Bashan Y, Gonzalez LE. 1999. Long-term survival of the plant-growth-promoting bacteria Azospirillum brasilense and Pseudomonas fluorescens in dry alginate inoculant. Applied Microbiology and Biotechnology. Feb;51:262-266.

Bautista-Cruz A, Donaji Ortiz-Hernandez Y, Martinez-Gallegos V, Martinez Gutierrez G. 2015. Effect of phosphate-solubilizing bacteria isolated from semiarid soils on pitahaya seedlings (Hylocereus undatus). Idesia. Mar-May;33:65-68.

Belimov AA, Dodd IC, Safronova VI, Shaposhnikov AI, Azarova TS, Makarova NM, Davies WJ, Tikhonovich IA. 2015. Rhizobacteria that produce auxins and contain 1-amino-cyclopropane-1-carboxylic acid deaminase decrease amino acid concentrations in the rhizosphere and improve growth and yield of well-watered and water-limited potato (Solanum tuberosum). Annals of Applied Biology. Jul;167:11-25.

Berg G, Smalla K. 2009. Plant species and soil type cooperatively shape the structure and function of microbial communities in the rhizosphere. Fems Microbiology Ecology. Apr;68:1-13.

Blakney AJC, Patten CL. 2011. A plant growth-promoting pseudomonad is closely related to the Pseudomonas syringae complex of plant pathogens. Fems Microbiology Ecology. Sep;77:546-557.

Bogino P, Nievas F, Banchio E, Giordano W. 2011. Increased competitiveness and efficiency of biological nitrogen fixation in peanut via in-furrow inoculation of rhizobia. European Journal of Soil Biology. May-Jun;47:188-193.

Boukhalfa H, Crumbliss AL. 2002. Chemical aspects of siderophore mediated iron transport. Biometals. Dec;15:325-339.

Castro-Sowinski S, Herschkovitz Y, Okon Y, Jurkevitch E. 2007. Effects of inoculation with plant growth-promoting rhizobacteria on resident rhizosphere microorganisms. Fems Microbiology Letters. Nov;276:1-11.

Chaudhry V, Rehman A, Mishra A, Chauhan PS, Nautiyal CS. 2012. Changes in Bacterial Community Structure of Agricultural Land Due to Long-Term Organic and Chemical Amendments. Microbial Ecology. Aug;64:450-460.

Chung EJ, Hossain MT, Khan A, Kim KH, Jeon CO, Chung YR. 2015. Bacillus oryzicola sp nov., an Endophytic Bacterium Isolated from the Roots of Rice with Antimicrobial, Plant Growth Promoting, and Systemic Resistance Inducing Activities in Rice. Plant Pathology Journal. Jun;31:152-164.

Costa JL, Paulsrud P, Lindblad P. 1999. Cyanobiont diversity within coralloid roots of selected cycad species. Fems Microbiology Ecology. Jan;28:85-91.

de la Cruz CPP, Bird CO, Isulat MD. 2012. Sprouting, Survival and Growth of Young Sugarcane (Saccharum officinarum L.) Treated with Diazotrophic Bacteria (Gluconacetobacter diazotrophicus). Philippine Agricultural Scientist. Mar;95:106-111.

de Souza R, Meyer J, Schoenfeld R, da Costa PB, Passaglia LMP. 2015. Characterization of plant growth-promoting bacteria associated with rice cropped in iron-stressed soils. Annals of Microbiology. Jun;65:951-964.

Debenport SJ, Assigbetse K, Bayala R, Chapuis-Lardy L, Dick RP, Gardener BBM. 2015. Association of Shifting Populations in the Root Zone Microbiome of Millet with Enhanced Crop Productivity in the Sahel Region (Africa). Applied and Environmental Microbiology. Apr;81:2841-2851.

Del Papa MF, Balague LJ, Sowinski SC, Wegener C, Segundo E, Abarca FM, Toro N, Niehaus K, Puhler A, Aguilar OM, et al. 1999. Isolation and characterization of alfalfa-nodulating rhizobia present in acidic soils of Central Argentina and Uruguay. Applied and Environmental Microbiology. Apr;65:1420-1427.

Diaz-Zorita M, Fernandez-Canigia MV. 2009. Field performance of a liquid formulation of Azospirillum brasilense on dryland wheat productivity. European Journal of Soil Biology. Jan-Feb;45:3-11.

Dong Z, Canny MJ, McCully ME, Roboredo MR, Cabadilla CF, Ortega E, Rodes R. 1994. A Nitrogen-Fixing Endophyte of Sugarcane Stems (A New Role for the Apoplast). Plant Physiology.105:1139-1147.

Drake JE, Darby BA, Giasson MA, Kramer MA, Phillips RP, Finzi AC. 2013. Stoichiometry constrains microbial response to root exudation-insights from a model and a field experiment in a temperate forest. Biogeosciences.10:821-838.

El-Katatny MH. 2010. Enzyme Production and Nitrogen Fixation by Free, Immobilized and Coimmobilized Inoculants of Trichoderma harzianum and Azospirillum brasilense and Their Possible Role in Growth Promotion of Tomato. Food Technology and Biotechnology. Apr-Jun;48:161-174.

Fedorova EE, Alzhapparova ZK, Zhiznevskaya GY, Artemenko EN, Izmailov SF. 1992. Phytohormones in soybean root-nodules. Soviet Plant Physiology. Mar-Apr;39:135-139.

Gagne-Bourgue F, Aliferis KA, Seguin P, Rani M, Samson R, Jabaji S. 2013. Isolation and characterization of indigenous endophytic bacteria associated with leaves of switchgrass (Panicum virgatum L.) cultivars. Journal of Applied Microbiology. Mar;114:836-853.

Gagne-Bourque F, Mayer BF, Charron JB, Vali H, Bertrand A, Jabaji S. 2015. Accelerated Growth Rate and Increased Drought Stress Resilience of the Model Grass Brachypodium distachyon Colonized by Bacillus subtilis B26. Plos One. Jun;10:23.

Gauri SS, Mandal SM, Pati BR. 2012. Impact of Azotobacter exopolysaccharides on sustainable agriculture. Applied Microbiology and Biotechnology. Jul;95:331-338.

Glick BR, Penrose DM, Li JP. 1998. A model for the lowering of plant ethylene concentrations by plant growth-promoting bacteria. Journal of Theoretical Biology. Jan;190:63-68.

Gohlke J, Deeken R. 2014. Plant responses to Agrobacterium tumefaciens and crown gall development. Frontiers in Plant Science. Apr;5:11.

Goncharova LI, Chizh TV, Murygin YV, Gubareva OS. 2010. Effect of Copper Contamination on the Growth and Biochemical Parameters of Field Bean Plants. Agrokhimiya. Dec:58-62.

Grichko VP, Glick BR. 2001. Amelioration of flooding stress by ACC deaminase-containing plant growth-promoting bacteria. Plant Physiology and Biochemistry. Jan;39:11-17.

Guo LN, Wu ZS, Rasool A, Li C. 2012. Effects of free and encapsulated co-culture bacteria on cotton growth and soil bacterial communities. European Journal of Soil Biology. Nov-Dec;53:16-22.

Han JH, Shim H, Shin JH, Kim KS. 2015. Antagonistic Activities of Bacillus spp. Strains Isolated from Tidal Flat Sediment Towards Anthracnose Pathogens Colletotrichum acutatum and C. gloeosporioides in South Korea. Plant Pathology Journal. Jun;31:165-175.

He YH, Wu ZS, Tu L, Han YJ, Zhang GL, Li C. 2015. Encapsulation and characterization of slow-release microbial fertilizer from the composites of bentonite and alginate. Applied Clay Science. Jun;109:68-75.

Hernandez RR, Debenport SJ, Leewis M, Ndoye F, Nkenmogne KIE, Soumare A, Thuita M, Gueye M, Miambi E, Chapuis-Lardy L, et al. 2015. The native shrub, Piliostigma reticulatum, as an ecological "resource island" for mango trees in the Sahel. Agriculture Ecosystems & Environment. Jun;204:51-61.

Hernandez-Leon R, Rojas-Solis D, Contreras-Perez M, Orozco-Mosqueda MD, Macias-Rodriguez LI, Reyes-de la Cruz H, Valencia-Cantero E, Santoyo G. 2015. Characterization of the antifungal and plant growth-promoting effects of diffusible and volatile organic compounds produced by Pseudomonas fluorescens strains. Biological Control. Feb;81:83-92.

Hien NT, Toan PV, Choudhury A, Rose MT, Roughley RJ, Kennedy IR. 2014. Field application strategies for the inoculant biofertilizer biogro supplementing fertilizer nitrogen application in rice production. Journal of Plant Nutrition.37:1837-1858.

Hoffman MT, Gunatilaka MK, Wijeratne K, Gunatilaka L, Arnold AE. 2013. Endohyphal Bacterium Enhances Production of Indole-3-Acetic Acid by a Foliar Fungal Endophyte. Plos One. Sep;8:8.

Hussain M, Jang KH, Farooq M, Lee DJ. 2012. Morphological and Physiological Evaluation of Korean Rice Genotypes for Salt Resistance. International Journal of Agriculture and Biology. Nov;14:970-974.

Hynes RK, Craig KA, Covert D, Smith RS, Rennie RJ. 1995. Liquid rhizobial inoculants for lentil and field pea. Journal of Production Agriculture. Oct-Dec;8:547-552.

Jeong S, Moon HS, Nam K. 2014. Enhanced uptake and translocation of arsenic in Cretan brake fern (Pteris cretica L.) through siderophorearsenic complex formation with an aid of rhizospheric bacterial activity. Journal of Hazardous Materials. Sep;280:536-543.

Jin CW, Du ST, Shamsi IH, Luo BF, Lin XY. 2011. NO synthase-generated NO acts downstream of auxin in regulating Fe-deficiency-induced root branching that enhances Fe-deficiency tolerance in tomato plants. Journal of Experimental Botany. Jul;62:3875-3884.

Junker RR, Tholl D. 2013. Volatile Organic Compound Mediated Interactions at the Plant-Microbe Interface. Journal of Chemical Ecology. Jul;39:810-825.

Karthik N, Binod P, Pandey A. 2015. Purification and characterisation of an acidic and antifungal chitinase produced by a Streptomyces sp. Bioresource Technology. Jul;188:195-201.

Kaur G, Reddy MS. 2015. Effects of Phosphate-Solubilizing Bacteria, Rock Phosphate and Chemical Fertilizers on Maize-Wheat Cropping Cycle and Economics. Pedosphere. Jun;25:428-437.

Khabbaz SE, Zhang L, Caceres LA, Sumarah M, Wang A, Abbasi PA. 2015. Characterisation of antagonistic Bacillus and Pseudomonas strains for biocontrol potential and suppression of damping-off and root rot diseases. Annals of Applied Biology. May;166:456-471.

Kopyra M, Gwozdz EA. 2004. The role of nitric oxide in plant growth regulation and responses to abiotic stresses. Acta Physiologiae Plantarum.26:459-472.

Koul A, Tripathi C, Adholeya A, Kochar M. 2015. Nitric oxide metabolism and indole acetic acid biosynthesis cross-talk in Azospirillum brasilense SM. Research in Microbiology. Apr;166:174-185.

Koul V, Adholeya A, Kochar M. 2015. Sphere of influence of indole acetic acid and nitric oxide in bacteria. Journal of Basic Microbiology. May;55:543-553.

Kudoyarova GR, Melentiev AI, Martynenko EV, Timergalina LN, Arkhipova TN, Shendel GV, Kuz'mina LY, Dodd IC, Veselov SY. 2014. Cytokinin producing bacteria stimulate amino acid deposition by wheat roots. Plant Physiology and Biochemistry. Oct;83:285-291.

Kumar V, Singh S, Singh J, Upadhyay N. 2015. Potential of Plant Growth Promoting Traits by Bacteria Isolated from Heavy Metal Contaminated Soils. Bulletin of Environmental Contamination and Toxicology. Jun;94:807-814.

Kusari P, Kusari S, Lamshoft M, Sezgin S, Spiteller M, Kayser O. 2014. Quorum quenching is an antivirulence strategy employed by endophytic bacteria. Applied Microbiology and Biotechnology. Aug;98:7173-7183.

Lin GH, Chang CY, Lin HR. 2015. Systematic profiling of indole-3-acetic acid biosynthesis in bacteria using LC-MS/MS. Journal of Chromatography B-Analytical Technologies in the Biomedical and Life Sciences. Apr;988:53-58.

Liu FC, Xing SJ, Ma HL, Du ZY, Ma BY. 2013. Cytokinin-producing, plant growth-promoting rhizobacteria that confer resistance to drought stress in Platycladus orientalis container seedlings. Applied Microbiology and Biotechnology. Oct;97:9155-9164.

Liu LL, Du WY, Luo WY, Su Y, Hui JJ, Ma SW. 2015a. Development of an Engineered Soil Bacterium Enabling to Convert Both Insoluble Inorganic and Organic Phosphate into Plant Available Phosphate and Its Use as a Biofertilizer. Molecular Biotechnology. May;57:419-429.

Liu YX, Li X, Cai K, Cai LT, Lu N, Shi JX. 2015b. Identification of benzoic acid and 3-phenylpropanoic acid in tobacco root exudates and their role in the growth of rhizosphere microorganisms. Applied Soil Ecology. Sep;93:78-87.

Lynch JM, Benedetti H, Insam H, Nuti MP, Smalla K, Torsvik V, Nannipieri P. 2004. Microbial diversity in soil: ecological theories, the contribution of molecular techniques and the impact of transgenic plants and transgenic microorganisms. Biology and Fertility of Soils. Oct:40:363-385.

Ma XM, Liu XX, Ning X, Zhang B, Han F, Guan XM, Tan YF, Zhang QW. 2008. Effects of Bacillus thuringiensis toxin Cry1Ac and Beauveria bassiana on Asiatic corn borer (Lepidoptera : Crambidae). Journal of Invertebrate Pathology. Oct;99:123-128.

Ma Y, Oliveira RS, Nai FJ, Rajkumar M, Luo YM, Rocha I, Freitas H. 2015. The hyperaccumulator Sedum plumbizincicola harbors metal-resistant endophytic bacteria that improve its phytoextraction capacity in multi-metal contaminated soil. Journal of Environmental Management. Jun;156:62-69.

Madhaiyan M, Poonguzhali S, Sa T. 2007. Characterization of 1-aminocyclopropane-1-carboxylate (ACC) deaminase containing Methylobacterium oryzae and interactions with auxins and ACC regulation of ethylene in canola (Brassica campestris). Planta. Sep;226:867-876.

Maheshwari DK, Dubey RC, Agarwal M, Dheeman S, Aeron A, Bajpai VK. 2015. Carrier based formulations of biocoenotic consortia of disease suppressive Pseudomonas aeruginosa KRP1 and Bacillus licheniformis KRB1. Ecological Engineering. Aug;81:272-277.

Maheshwari DK, Kumar S, Kumar B, Pandey P. 2010. Co-inoculation of Urea and DAP Tolerant Sinorhizobium meliloti and Pseudomonas aeruginosa as Integrated Approach for Growth Enhancement of Brassica juncea. Indian Journal of Microbiology. Oct;50:425-431.

Mahgoob AEA, El-Tayeb TS. 2010. Biological Control of the Root-Knot Nematode, Meloidogyne incognita on Tomato Using Plant Growth Promoting Bacteria. Egyptian Journal of Biological Pest Control.20:95-103.

Maindad DV, Kasture VM, Chaudhari H, Dhavale DD, Chopade BA, Sachdev DP. 2014. Characterization and Fungal Inhibition Activity of Siderophore from Wheat Rhizosphere Associated Acinetobacter calcoaceticus Strain HIRFA32. Indian Journal of Microbiology. Sep;54:315-322.

Marchetti M, Capela D, Glew M, Cruveiller S, Chane-Woon-Ming B, Gris C, Timmers T, Poinsot V, Gilbert LB, Heeb P, et al. 2010. Experimental Evolution of a Plant Pathogen into a Legume Symbiont. Plos Biology. Jan;8:10.

Matraszek R, Hawrylak-Nowak B. 2010. Growth and mineral composition of nickel-stressed plants under conditions of supplementation with excessive amounts of calcium and iron. Journal of Toxicology and Environmental Health-Part a-Current Issues.73:1260-1273.

Medeiros AFA, Polidoro JC, Reis VM. 2006. Nitrogen source effect on Gluconacetobacter diazotrophicus colonization of sugarcane (Saccharum spp.). Plant and Soil. Feb;279:141-152.

Menesguen A, Dion P. 2009. Role of phosphorus in coastal eutrophication. Oceanis, Vol 33, No 1 and 2.33:17-35.

Miller G, Suzuki N, Ciftci-Yilmaz S, Mittler R. 2010. Reactive oxygen species homeostasis and signalling during drought and salinity stresses. Plant Cell and Environment. Apr;33:453-467.

Mishra PK, Bisht SC, Ruwari P, Joshi GK, Singh G, Bisht JK, Bhatt JC. 2011. Bioassociative effect of cold tolerant Pseudomonas spp. and Rhizobium leguminosarum-PR1 on iron acquisition, nutrient uptake and growth of lentil (Lens culinaris L.). European Journal of Soil Biology. Jan-Feb;47:35-43.

Muller D, Waldie T, Miyawaki K, To JPC, Melnyk CW, Kieber JJ, Kakimoto T, Leyser O. 2015. Cytokinin is required for escape but not release from auxin mediated apical dominance. Plant Journal. Jun;82:874-886.

Nagata S, Yamaji K, Nomura N, Ishimoto H. 2015. Root endophytes enhance stress-tolerance of Cicuta virosa L. growing in a mining pond of eastern Japan. Plant Species Biology. Apr;30:116-125.

Nakaew N, Rangjaroen C, Sungthong R. 2015. Utilization of rhizospheric Streptomyces for biological control of Rigidoporus sp causing white root disease in rubber tree. European Journal of Plant Pathology. May;142:93-105.

Ndungu-Magiroi KW, Waswa B, Bationo A, Okalebo JR, Othieno C, Herrmann L, Lesueur D. 2015. Minjingu phosphate rock applications increase the population of phosphate solubilising microorganisms with a positive impact on crop yields in a Kenyan Ferralsol. Nutrient Cycling in Agroecosystems. May;102:91-99.

Oliveira ALM, Stoffels M, Schmid M, Reis VM, Baldani JI, Hartmann A. 2009. Colonization of sugarcane plantlets by mixed inoculations with diazotrophic bacteria. European Journal of Soil Biology. Jan-Feb;45:106-113.

Ona O, Van Impe J, Prinsen E, Vanderleyden J. 2005. Growth and indole-3-acetic acid biosynthesis of Azospirillum brasilense Sp245 is environmentally controlled. Fems Microbiology Letters. May;246:125-132.

Pagnussat GC, Simontacchi M, Puntarulo S, Lamattina L. 2002. Nitric oxide is required for root organogenesis. Plant Physiology. Jul;129:954-956.

Park YS, Dutta S, Ann M, Raaijmakers JM, Park K. 2015. Promotion of plant growth by Pseudomonas fluorescens strain SS101 via novel volatile organic compounds. Biochemical and Biophysical Research Communications. May;461:361-365.

Patten CL, Blakney AJC, Coulson TJD. 2013. Activity, distribution and function of indole-3-acetic acid biosynthetic pathways in bacteria. Critical Reviews in Microbiology. Nov;39:395-415.

Pedraza RO. 2008. Recent advances in nitrogen-fixing acetic acid bacteria. International Journal of Food Microbiology. Jun;125:25-35.

Pereira SIA, Barbosa L, Castro PML. 2015. Rhizobacteria isolated from a metal-polluted area enhance plant growth in zinc and cadmium-contaminated soil. International Journal of Environmental Science and Technology. Jul;12:2127-2142.

Radzki W, Manero FJG, Algar E, Garcia JAL, Garcia-Villaraco A, Solano BR. 2013. Bacterial siderophores efficiently provide iron to iron-starved tomato plants in hydroponics culture. Antonie Van Leeuwenhoek International Journal of General and Molecular Microbiology. Sep;104:321-330.

Rajkumar M, Lee KJ, Lee WH, Banu JR. 2005. Growth of Brassica juncea under chromium stress: Influence of siderophores and indole 3 acetic acid producing rhizosphere bacteria. Journal of Environmental Biology. Oct;26:693-699.

Rekha PD, Lai WA, Arun AB, Young CC. 2007. Effect of free and encapsulated Pseudomonas putida CC-FR2-4 and Bacillus subtilis CC-pg104 on plant growth under gnotoblotic conditions. Bioresource Technology. Jan;98:447-451.

Rodriguez H, Fraga R, Gonzalez T, Bashan Y. 2006. Genetics of phosphate solubilization and its potential applications for improving plant growth-promoting bacteria. Plant and Soil. Sep;287:15-21.

Ruiz JA, Bernar EM, Jung K. 2015. Production of Siderophores Increases Resistance to Fusaric Acid in Pseudomonas protegens Pf-5. Plos One. Jan;10:16.

Sachs JL, Skophammer RG, Bansal N, Stajich JE. 2014. Evolutionary origins and diversification of proteobacterial mutualists. Proceedings of the Royal Society B-Biological Sciences. Jan;281:9.

Saleem MM, Arshad M, Yaseen M. 2013. Effectiveness of Various Approaches to Use Rock Phosphate as a Potential Source of Plant Available P for Sustainable Wheat Production. International Journal of Agriculture and Biology.15:223-230.

Sanz L, Albertos P, Mateos I, Sanchez-Vicente I, Lechon T, Fernandez-Marcos M, Lorenzo O. 2015. Nitric oxide (NO) and phytohormones crosstalk during early plant development. Journal of Experimental Botany. May;66:2857-2868.

Saravanan VS, Madhaiyan M, Osborne J, Thangaraju M, Sa TM. 2008. Ecological occurrence of Gluconacetobacter diazotrophicus and nitrogen-fixing Acetobacteraceae members: Their possible role in plant growth promotion. Microbial Ecology. Jan;55:130-140.

Schmulling T. 2002. New insights into the functions of cytokinins in plant development. Journal of Plant Growth Regulation. Mar;21:40-49.

Sethia B, Mustafa M, Manohar S, Patil SV, Jayamohan NS, Kumudini BS. 2015. Indole acetic acid production by fluorescent Pseudomonas spp. from the rhizosphere of Plectranthus amboinicus (Lour.) Spreng. and their variation in extragenic repetitive DNA sequences. Indian Journal of Experimental Biology. Jun;53:342-349.

Shaikh SS, Patel PR, Patel SS, Nikam SD, Rane TU, Sayyed RZ. 2014. Production of biocontrol traits by banana field fluorescent Pseudomonads and comparison with chemical fungicide. Indian Journal of Experimental Biology. Sep;52:917-920.

Shapiro AD. 2005. Nitric oxide signaling in plants. In: Plant Hormones. San Diego: Elsevier Academic Press Inc. p. 339-398.

Shi H-T, Li R-J, Cai W, Liu W, Fu Z-W, Lu Y-T. 2012. In vivo role of nitric oxide in plant response to abiotic and biotic stress. Plant signaling & behavior. 2012 Mar (Epub 2012 Mar;7:437-439.

Siddiqui MH, Al-Whaibi MH, Basalah MO. 2011. Role of nitric oxide in tolerance of plants to abiotic stress. Protoplasma. Jul;248:447-455.

Singh RK, Suman A, Singh J, Singh AK, Kumar S, Singh PK. 2012. Effects of Gluconacetobacter diazotrophicus on seed cane produced through micro-propagated plantlets in sugarcane (Saccharum spp hybrid complex) under sub-tropics. Indian Journal of Agricultural Sciences. Apr;82:340-344.

Sousa NR, Franco AR, Ramos MA, Oliveira RS, Castro PML. 2015. The response of Betula pubescens to inoculation with an ectomycorrhizal fungus and a plant growth promoting bacterium is substrate-dependent. Ecological Engineering. Aug;81:439-443.

Subramanian P, Kim K, Krishnamoorthy R, Sundaram S, Sa TM. 2015a. Endophytic bacteria improve nodule function and plant nitrogen in soybean on co-inoculation with Bradyrhizobium japonicum MN110. Plant Growth Regulation. Jul;76:327-332.

Subramanian P, Krishnamoorthy R, Chanratana M, Kim K, Sa T. 2015b. Expression of an exogenous 1-aminocyclopropane-1-carboxylate deaminase gene in psychrotolerant bacteria modulates ethylene metabolism and cold induced genes in tomato under chilling stress. Plant Physiology and Biochemistry. Apr;89:18-23.

Sulochana MB, Jayachandra SY, Kumar SA, Parameshwar AB, Reddy KM, Dayanand A. 2014. Siderophore as a Potential Plant Growth-Promoting Agent Produced by Pseudomonas aeruginosa JAS-25. Applied Biochemistry and Biotechnology. Sep;174:297-308.

Takei T, Yoshida M, Hatate Y, Shiomori K, Kiyoyama S. 2008. Lactic Acid Bacteria-Enclosing Poly(epsilon-Caprolactone) Microcapsules as Soil Bioamendment. Journal of Bioscience and Bioengineering. Sep;106:268-272.

Tan H, Barret M, Rice O, Dowling DN, Burke J, Morrissey JP, O'Gara F. 2012. Long-term agrichemical use leads to alterations in bacterial community diversity. Plant Soil and Environment. Oct;58:452-458.

Tanaka S, Han XW, Kahmann R. 2015. Microbial effectors target multiple steps in the salicylic acid production and signaling pathway. Frontiers in Plant Science. May;6:10.

Tank N, Saraf M. 2009. Enhancement of plant growth and decontamination of nickel-spiked soil using PGPR. Journal of Basic Microbiology. Apr;49:195-204.

Tavares DA, Roat TC, Carvalho SM, Silva-Zacarin ECM, Malaspina O. 2015. In vitro effects of thiamethoxam on larvae of Africanized honey bee Apis mellifera (Hymenoptera: Apidae). Chemosphere. Sep;135:370-378.

Tomic JM, Milivojevic JM, Pesakovic MI. 2015. The response to bacterial inoculation is cultivar-related in strawberries. Turkish Journal of Agriculture and Forestry.39:332-341.

van Olden C, Groen AK, Nieuwdorp M. 2015. Role of Intestinal Microbiome in Lipid and Glucose Metabolism in Diabetes Mellitus. Clinical Therapeutics. Jun;37:1172-1177.

Vassilev N, Vassileva M, Lopez A, Martos V, Reyes A, Maksimovic I, Eichler-Lobermann B, Malusa E. 2015. Unexploited potential of some biotechnological techniques for biofertilizer production and formulation. Applied Microbiology and Biotechnology. Jun;99:4983-4996.

Viruel E, Erazzu LE, Calsina LM, Ferrero MA, Lucca ME, Sineriz F. 2015. Inoculation of maize with phosphate solubilizing bacteria: effect on plant growth and yield. Journal of Soil Science and Plant Nutrition.14:819-831.

Wang JF, Zhang YQ, Li Y, Wang XM, Nan WB, Hu YF, Zhang H, Zhao CZ, Wang F, Li P, et al. 2015a. Endophytic microbes Bacillus sp LZR216-regulated root development is dependent on polar auxin transport in Arabidopsis seedlings. Plant Cell Reports. Jun;34:1075-1087.

Wang JJ, Zhang HW, Zhang XL, Qin SH, Tan HB, Li XY. 2013. Effects of long-term chlorimuron-ethyl application on the diversity and antifungal activity of soil Pseudomonas spp. in a soybean field in Northeast China. Annals of Microbiology. Mar;63:335-341.

Wang XM, Yang B, Ren CG, Wang HW, Wang JY, Dai CC. 2015b. Involvement of abscisic acid and salicylic acid in signal cascade regulating bacterial endophyte-induced volatile oil biosynthesis in plantlets of Atractylodes lancea. Physiologia Plantarum. Jan;153:30-42.

Wu CS. 2008. Controlled release evaluation of bacterial fertilizer using polymer composites as matrix. Journal of Controlled Release. Nov;132:42-48.

Wu KJ, Wu CS, Chang JS. 2007. Biodegradability and mechanical properties of polycaprolactone composites encapsulating phosphate-solubilizing bacterium Bacillus sp PG01. Process Biochemistry. Apr;42:669-675.

Wu ZS, Guo LN, Qin SH, Li C. 2012. Encapsulation of R. planticola Rs-2 from alginate-starch-bentonite and its controlled release and swelling behavior under simulated soil conditions. Journal of Industrial Microbiology & Biotechnology. Feb;39:317-327.

Wu ZS, Zhao YF, Kaleem I, Li C. 2011. Preparation of calcium-alginate microcapsuled microbial fertilizer coating Klebsiella oxytoca Rs-5 and its performance under salinity stress. European Journal of Soil Biology. Mar-Apr;47:152-159.

Xia XJ, Zhou YH, Shi K, Zhou J, Foyer CH, Yu JQ. 2015. Interplay between reactive oxygen species and hormones in the control of plant development and stress tolerance. Journal of Experimental Botany. May;66:2839-2856.

Xiao CQ, Fang YJ, Chi R. 2015. Phosphate solubilization in vitro by isolated Aspergillus niger and Aspergillus carbonarius. Research on Chemical Intermediates. May;41:2867-2878.

Yadegari M, Rahmani HA, Noormohammadi G, Ayneband A. 2010. Plant growth promoting rhizobacteria increase growth, yield and nitrogen fixation in phaseolus vulgaris. Journal of Plant Nutrition.33:1733-1743.

Yamasaki H. 2000. Nitrite-dependent nitric oxide production pathway: implications for involvement of active nitrogen species in photoinhibition in vivo. Philosophical Transactions of the Royal Society B-Biological Sciences. Oct;355:1477-1488.

Yandigeri MS, Malviya N, Solanki MK, Shrivastava P, Sivakumar G. 2015. Chitinolytic Streptomyces vinaceusdrappus S5MW2 isolated from Chilika lake, India enhances plant growth and biocontrol efficacy through chitin supplementation against Rhizoctonia solani. World Journal of Microbiology & Biotechnology. Aug;31:1217-1225.

Yang RX, Luo CL, Chen YH, Wang GP, Xu Y, Shen ZG. 2013. Copper-Resistant Bacteria Enhance Plant Growth and Copper Phytoextraction. International Journal of Phytoremediation. Jul;15:573-584.

Yarullina DR, Asafova EV, Kartunova JE, Ziyatdinova GK, Ilinskaya ON. 2014. Probiotics for plants: NO-producing lactobacilli protect plants from drought. Applied Biochemistry and Microbiology. Mar;50:166-168.

Young CC, Rekha P, Lai WA, Arun AB. 2006. Encapsulation of plant growth-promoting bacteria in alginate beads enriched with humic acid. Biotechnology and Bioengineering. Sep;95:76-83.

Yu X, Liu X, Zhu TH, Liu GH, Mao C. 2012. Co-inoculation with phosphate-solubilzing and nitrogen-fixing bacteria on solubilization of rock phosphate and their effect on growth promotion and nutrient uptake by walnut. European Journal of Soil Biology. May-Jun;50:112-117.

Zafar-ul-Hye M, Farooq HM, Hussain M. 2015. Bacteria in combination with fertilizers promote root and shoot growth of maize in saline-sodic soil. Brazilian Journal of Microbiology. May;46:97-102.

Zhang L, Khabbaz SE, Wang A, Li H, Abbasi PA. 2015. Detection and characterization of broad-spectrum antipathogen activity of novel rhizobacterial isolates and suppression of Fusarium crown and root rot disease of tomato. Journal of Applied Microbiology. Mar;118:685-703.

Zhao J, Xue QH, Niu GG, Xue L, Shen GH, Du JZ. 2013. Extracellular enzyme production and fungal mycelia degradation of antagonistic Streptomyces induced by fungal mycelia preparation of cucurbit plant pathogens. Annals of Microbiology. Jun;63:809-812.

Zilli JE, Gianluppi V, Campo RJ, Rouws JRC, Hungria M. 2010. In-furrow inoculation with bradyrhizobium alternatively to seed inoculation of soybean. Revista Brasileira De Ciencia Do Solo. Nov-Dec;34:1875-1881.

Vita

As a former field hand on an organic farm, my interests in botany have remained closely tied to the development of practices which can reduce our effect on the natural world. I've seen high yield and the proliferation of a variety of crops without the use of agricultural chemicals, this is already a reality for many farms using only organic sources of nutrient addition and pest management. The further development of inoculation could do a great deal of good, both for smaller scale and industrial scale agriculture.

Permanent email: mebels1@utexas.edu

This report was typed by Marcus Ebels.